15-July-2009

Dr. William S. Stokes, Director, NICEATM, NIEHS,
P.O. Box 12233,
Mail Stop: K2–16,
Research Triangle Park, NC 27709

Re: **Independent Scientific Peer Review Panel Report: Updated Validation Status of New Versions and Applications of the Murine Local Lymph Node Assay: A Test Method for Assessing the Allergic Contact Dermatitis Potential of Chemicals and Products**

Dear Dr. Stokes,

Sanofi-aventis U.S. Inc, a member of the sanofi-aventis Group, appreciates the opportunity to comment on the above-referenced report, the *Independent Scientific Peer Review Panel Report: Updated Validation Status of New Versions and Applications of the Murine Local Lymph Node Assay: A Test Method for Assessing the Allergic Contact Dermatitis Potential of Chemicals and Products* and provide the following comments:

General Comments

The document is quite technical and comments will focus on sections 1-3 and section 4, testing of pesticide formulation. Sanofi-aventis acknowledges some positive approaches to the LLNA methods proposed within the report. These approaches include the reduction in the number of animals, the replacement of the guinea pig, and the avoidance of radioactive compounds, and the use of negative and positive controls for the three methodologies. While the report offers three modified methodologies for the LLNA, these methodologies do not highlight significant progress from the classical LLNA.

Specific Comments

Section 1.0 – LLNA-DA

1) In this protocol the justification for replacing the guinea pig is provided. The replacement is not mentioned for the LLNA-BrdU-FC or the LLNA-BrdU-ELISA. It could be mentioned for the other two methodologies.

2) An explanation of the use of sodium lauryl sulfate is need due to ethical reasons.

Section 2.0 – LLNA BrdU-FC

1) In this protocol the ear swelling is recommended to evaluation irritancy. The assessment would be interesting for the LLNA-DA and LLNA BrdU-ELISA or the rationale to incorporate the ear swelling in this method needs to be explained.

2) The difficulties of the LLNA reside in classifying compound based on decision criteria for stimulation index and in discriminating irritancy from sensitization. The LLNA BrdU-FC method might offer the ability to discriminate irritants from sensitizers but might be problematic for weak sensitizers. For this assay, no inter-laboratory studies have been performed so a great deal of work is necessary to validate this approach.

Section 3.0 – LLNA BrdU-ELISA

1) The number of animals is not homogeneous between the three methodologies (LLNA-DA: 4 mice; LLNA BrDU-FC: 4-5 mice; LLNA BrdU-ELISA: 8 mice). The inconsistency might trigger the preference to avoid LLNA-BrdU-ELISA for ethical reason.

2) The validated benchmark for positive effect in the LLNA is a stimulation index of ≥ 3. When a value very close to 3 is observed, standard practice is to repeat the assay to obtain either a definitive result or confirm an equivocal finding. As written, the recommendation by ICCVAM appears to discourage this practice when using the LLNA BrdU-FC. This does not appear to be related to the number of animals needed and therefore there is no obvious explanation.

Sanofi-aventis appreciates the opportunity to comment on the draft ICCVAM report and hopes the comments provided are useful in preparing the final report.

Sincerely,

/s/

Brian E. Harvey, M.D., Ph.D.
Vice President
Regulatory Policy